#5

Local DNA Opening by PNA Creates Activated Nucleation Site for Both RecA-coated Complementary Single-stranded Probes

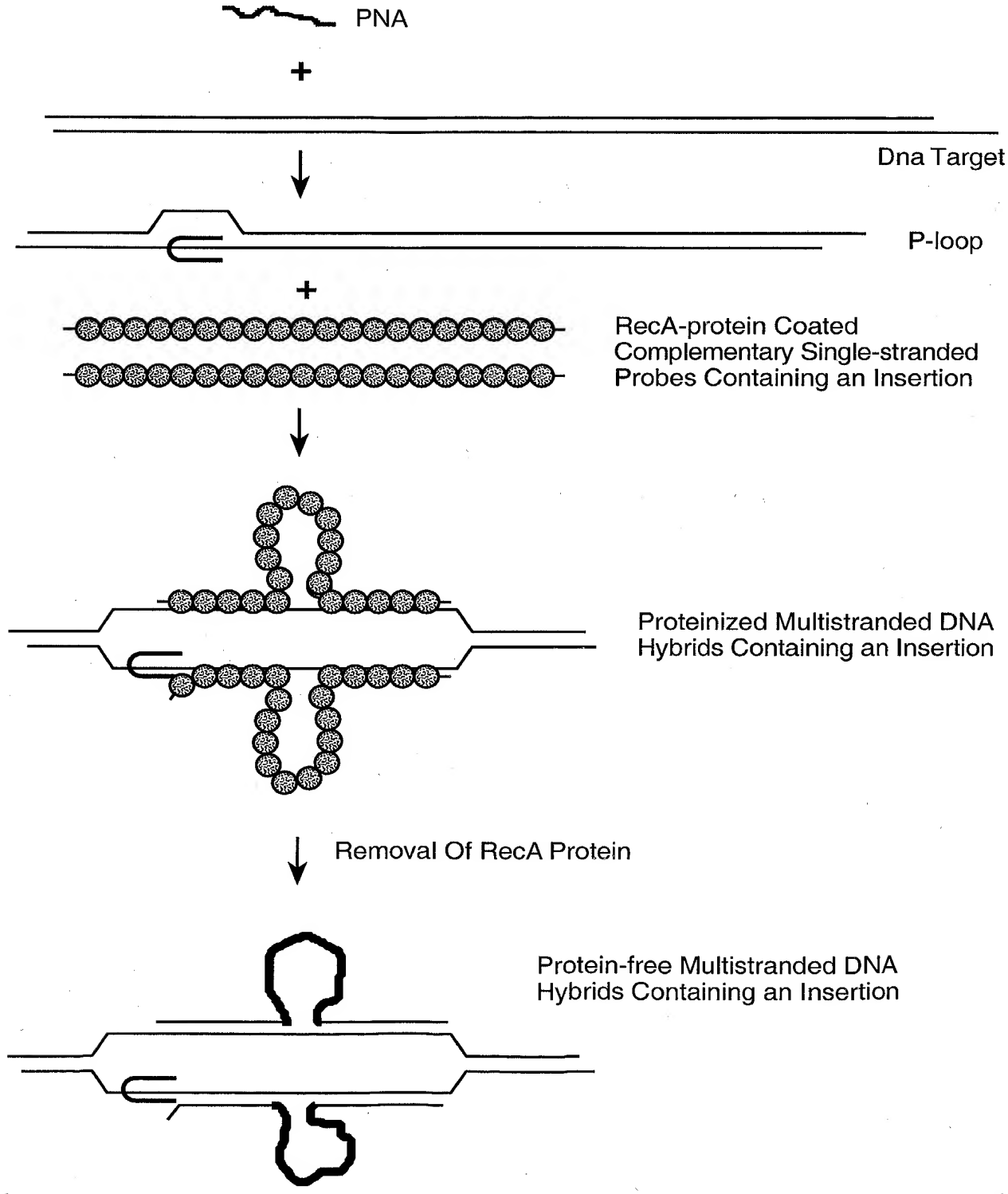

FIG._1A

Stabilization of Single D-loop Hybrids by Analog Probes:

When the PNA Binding Site is Within the Probe-target Hybrid, PNA Can Stabilize Single D-loops by Trapping the Strand Exchange Process.

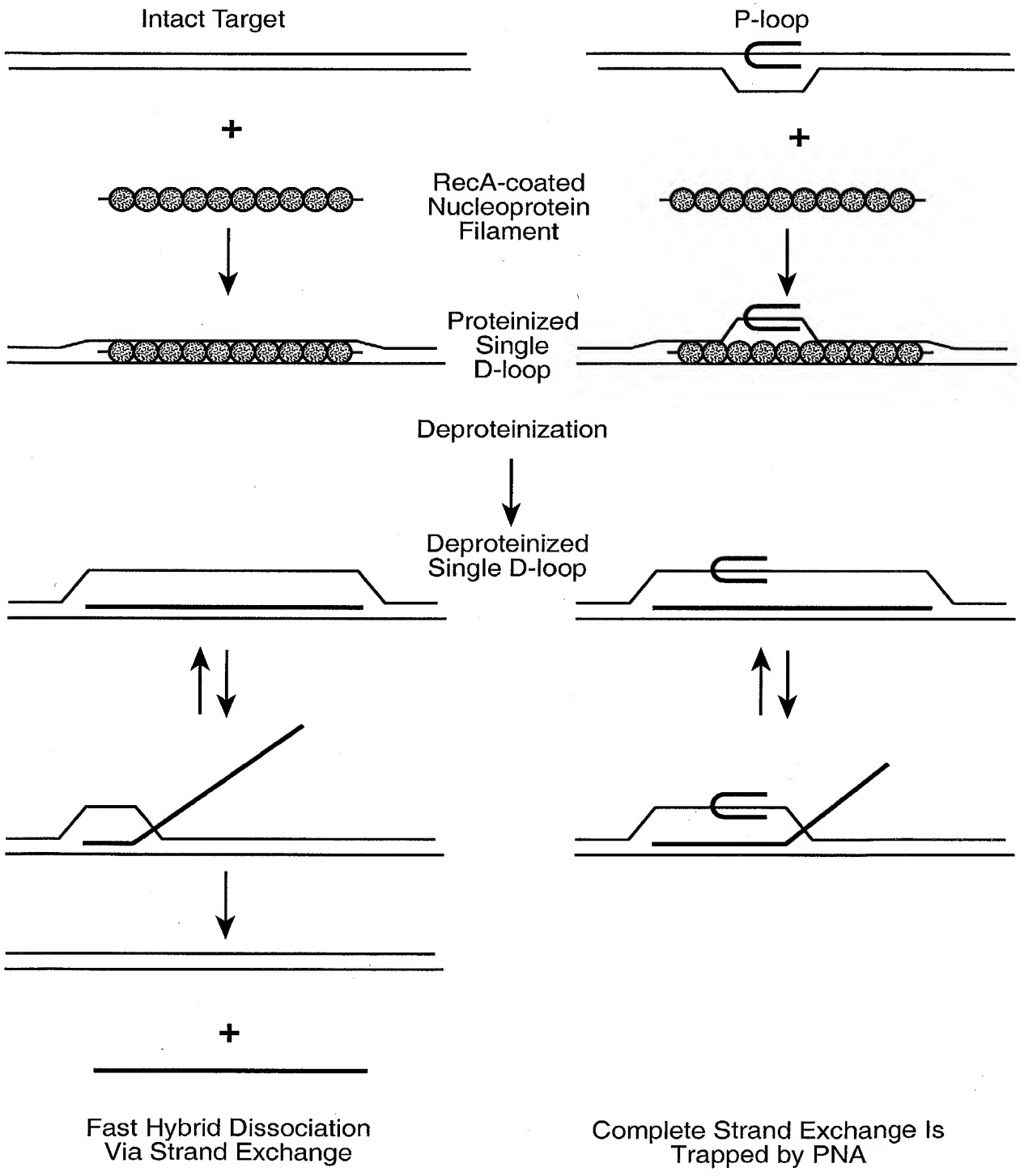

FIG._1B

PNA Activates the Binding of the Second Coming RecA-coated Single-stranded Probe Via Stabilization of the "Opened" State of the D-loop Formed by the First Coming RecA-coated Single-Stranded Probe.

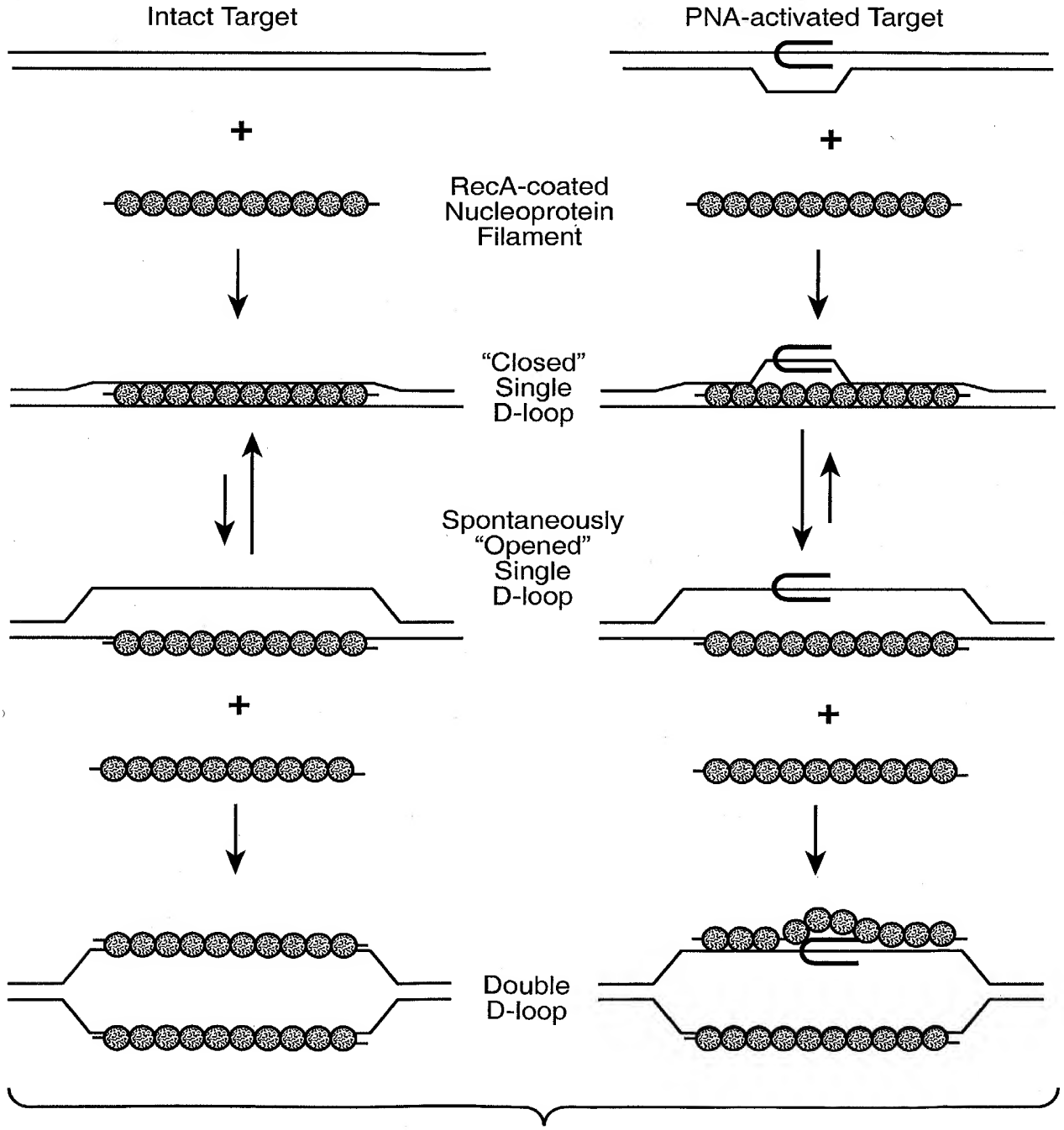

FIG._1C

PNA-directed Double-stranded Break In The Target DNA Followed By Homologous DNA Targeting

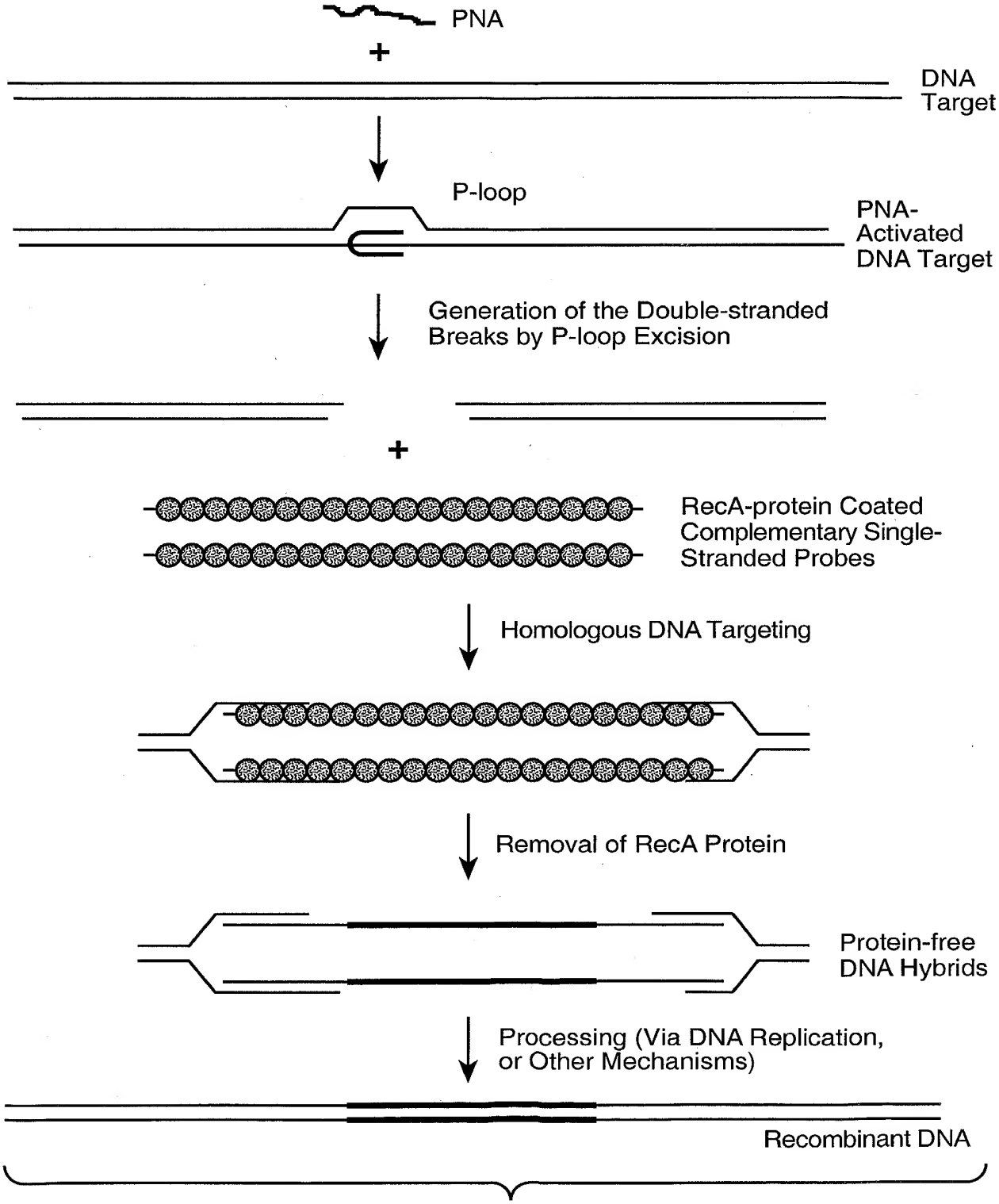

FIG._2

Processing of the Hybrids by Strand Excision Followed by DNA Repair

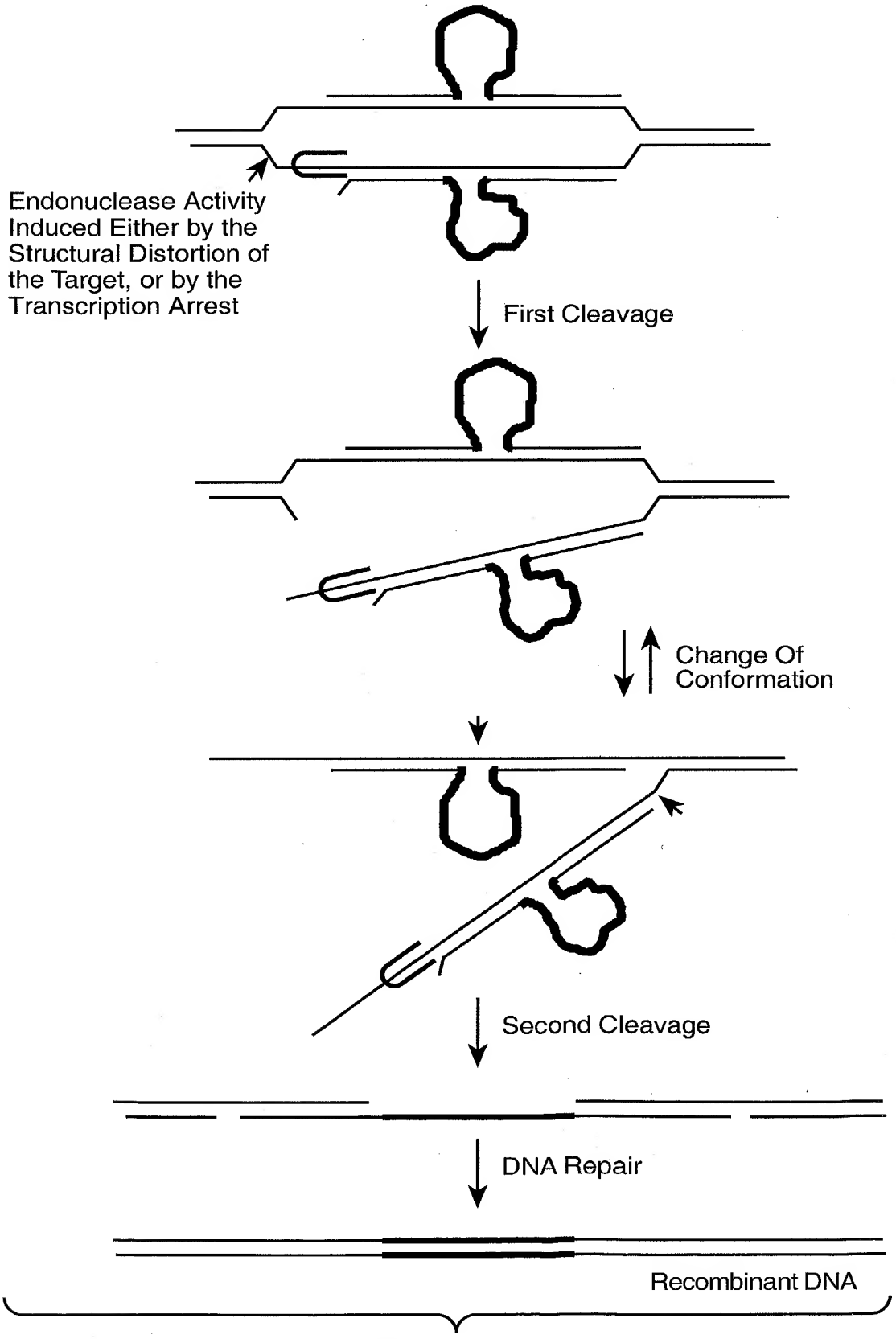

FIG._3A

Hybrid Processing Mediated by Target DNA Replication when the PNA Site is Outside the Heterologous Insert Site

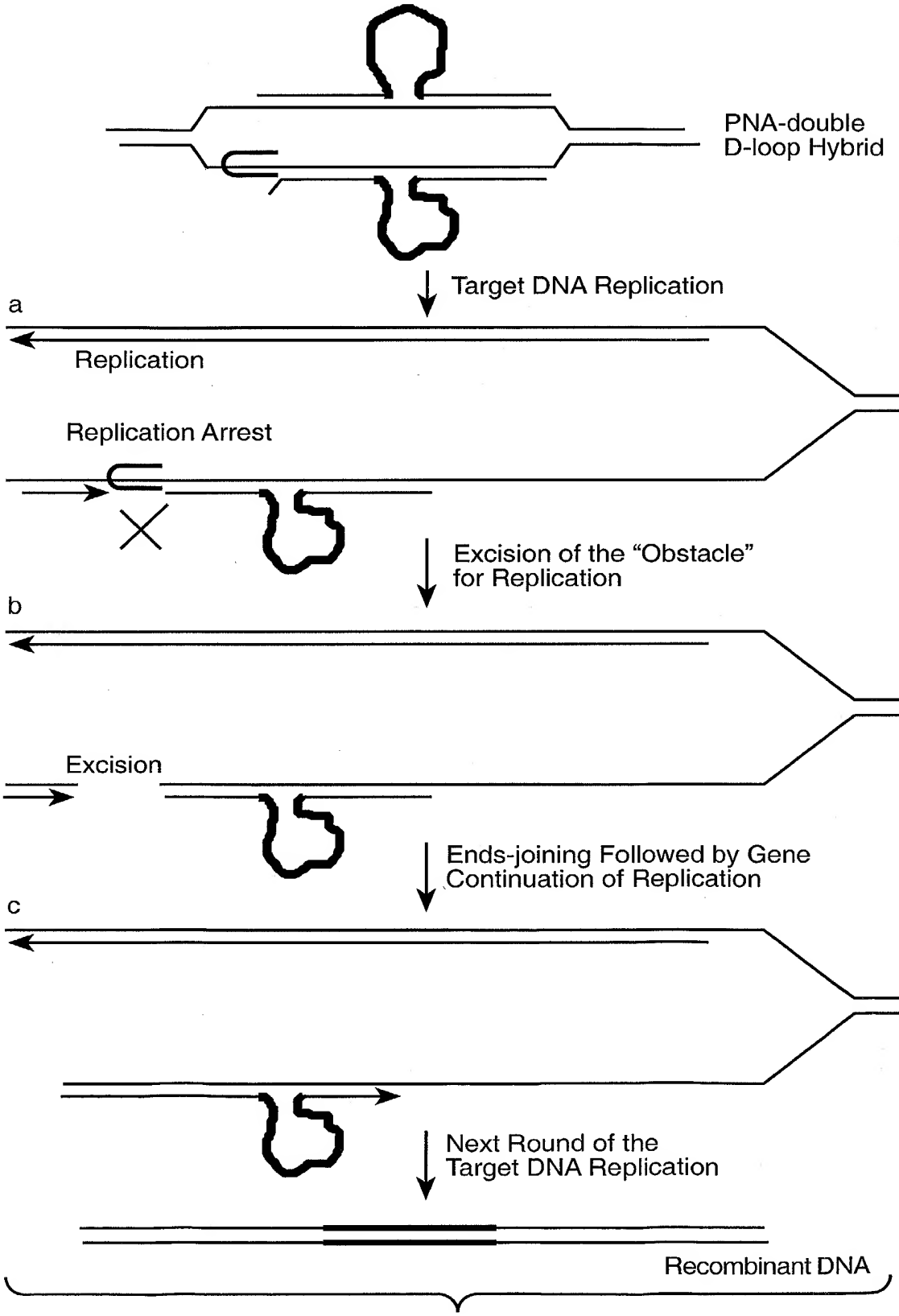

FIG._3B

Hybrid Processing Mediated by Target DNA Replication when the PNA Site is Inside the Heterologous Insert Site

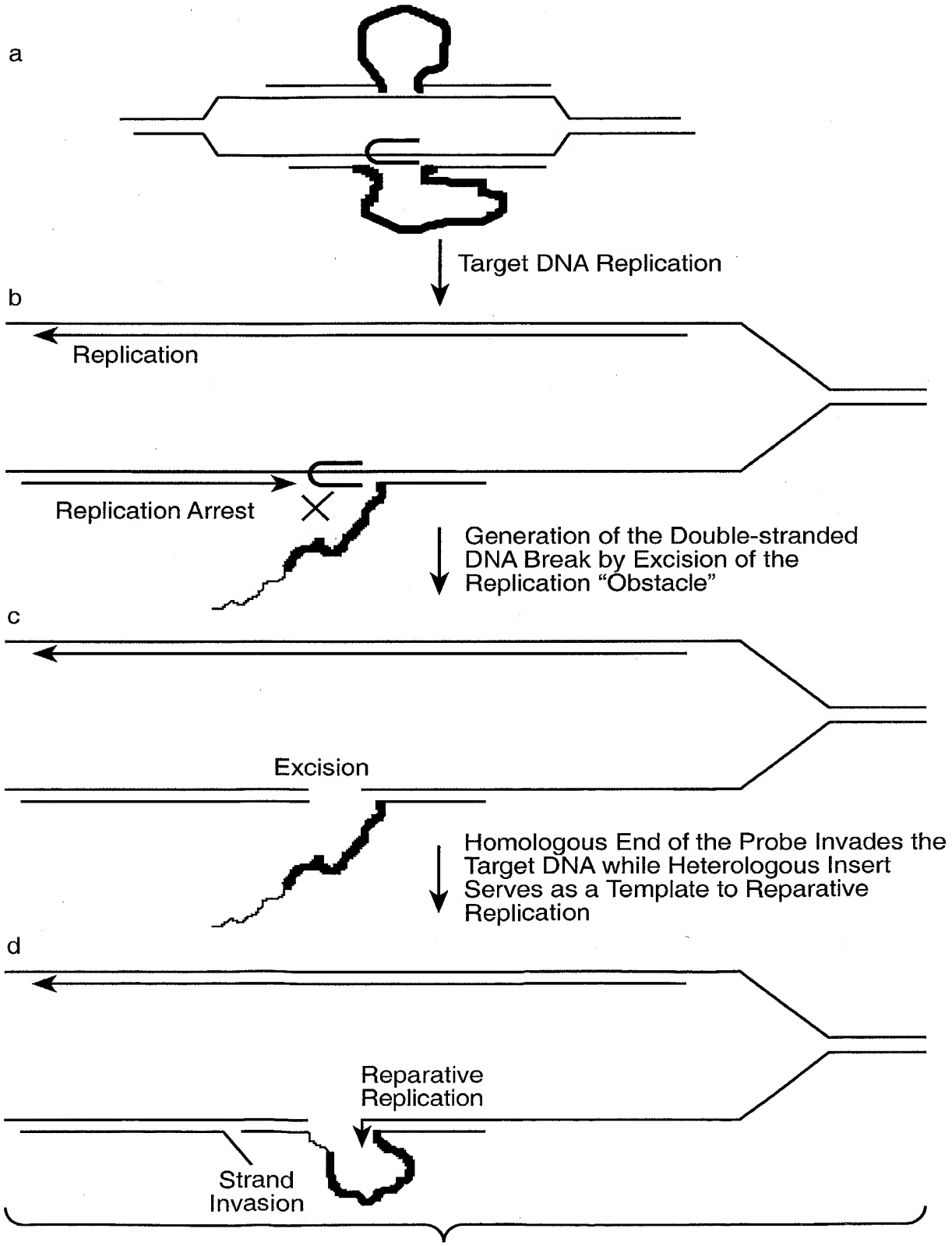

FIG._3C-1

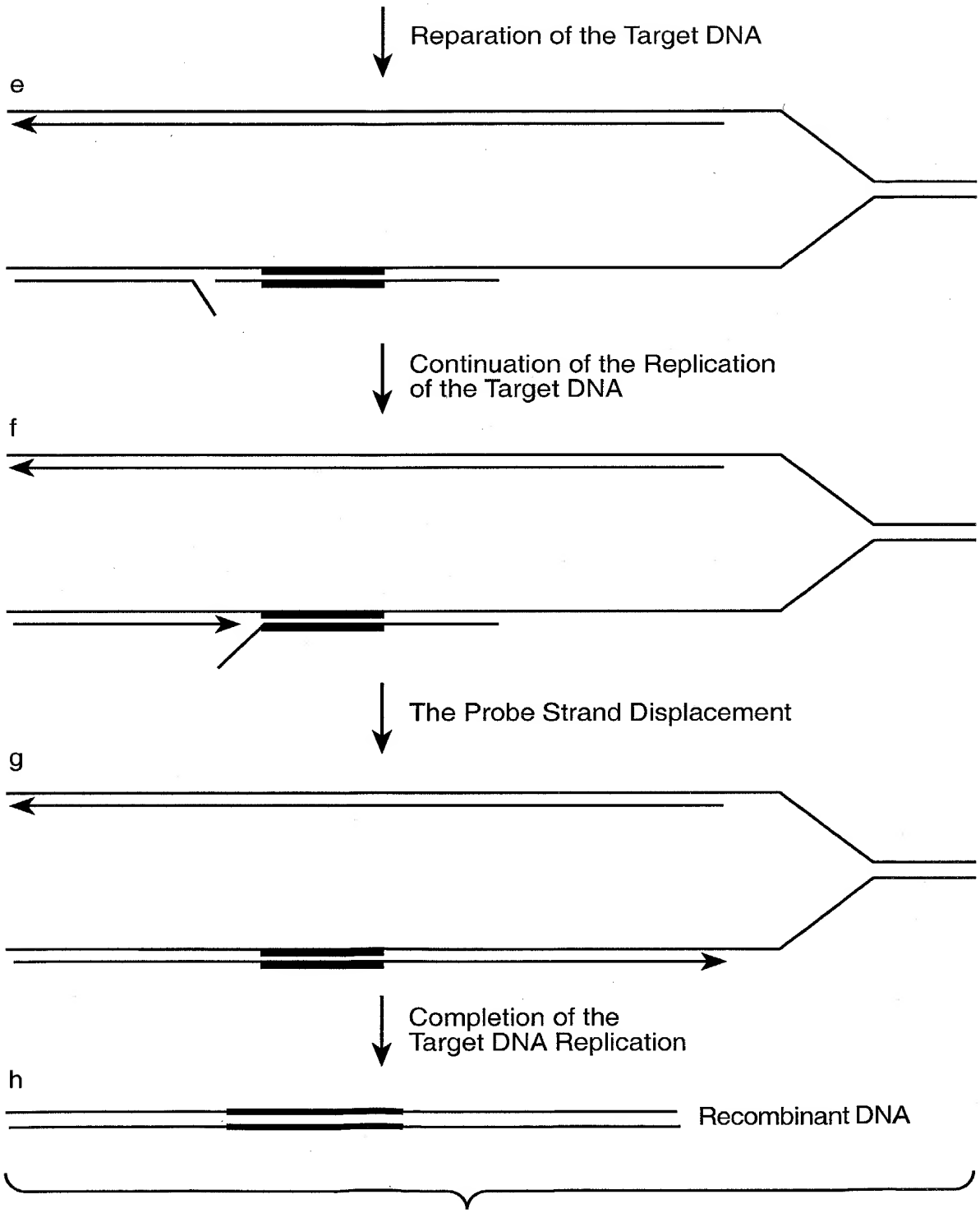
Reparation of the Target DNA
e
Continuation of the Replication
of the Target DNA
f
The Probe Strand Displacement
g
Completion of the
Target DNA Replication
h
Recombinant DNA

FIG._3C-2

Cloning of Linear (Including Genomic) DNA Fragments Mediated by PNA Activated Homologous DNA Targeting

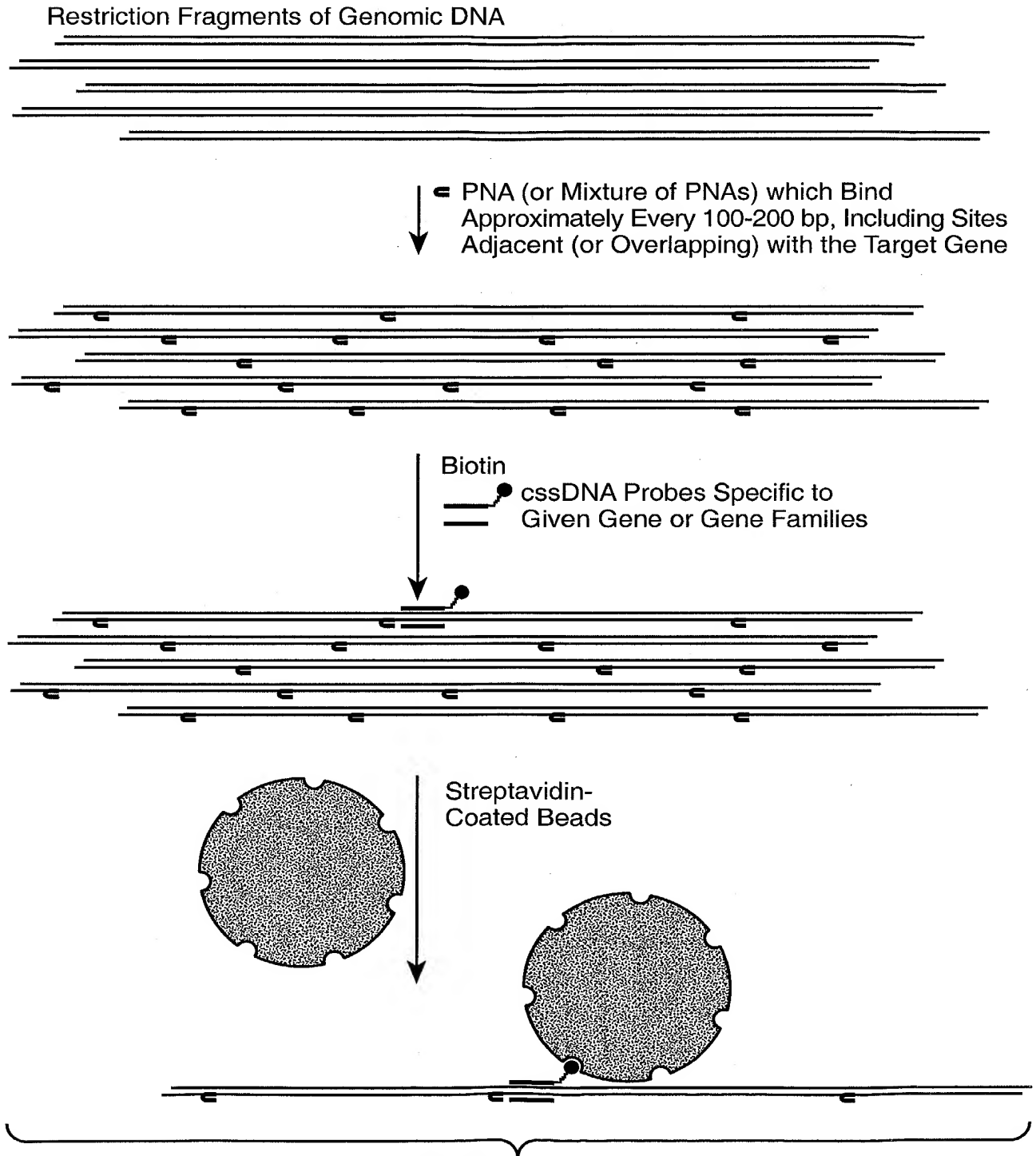

FIG._4A

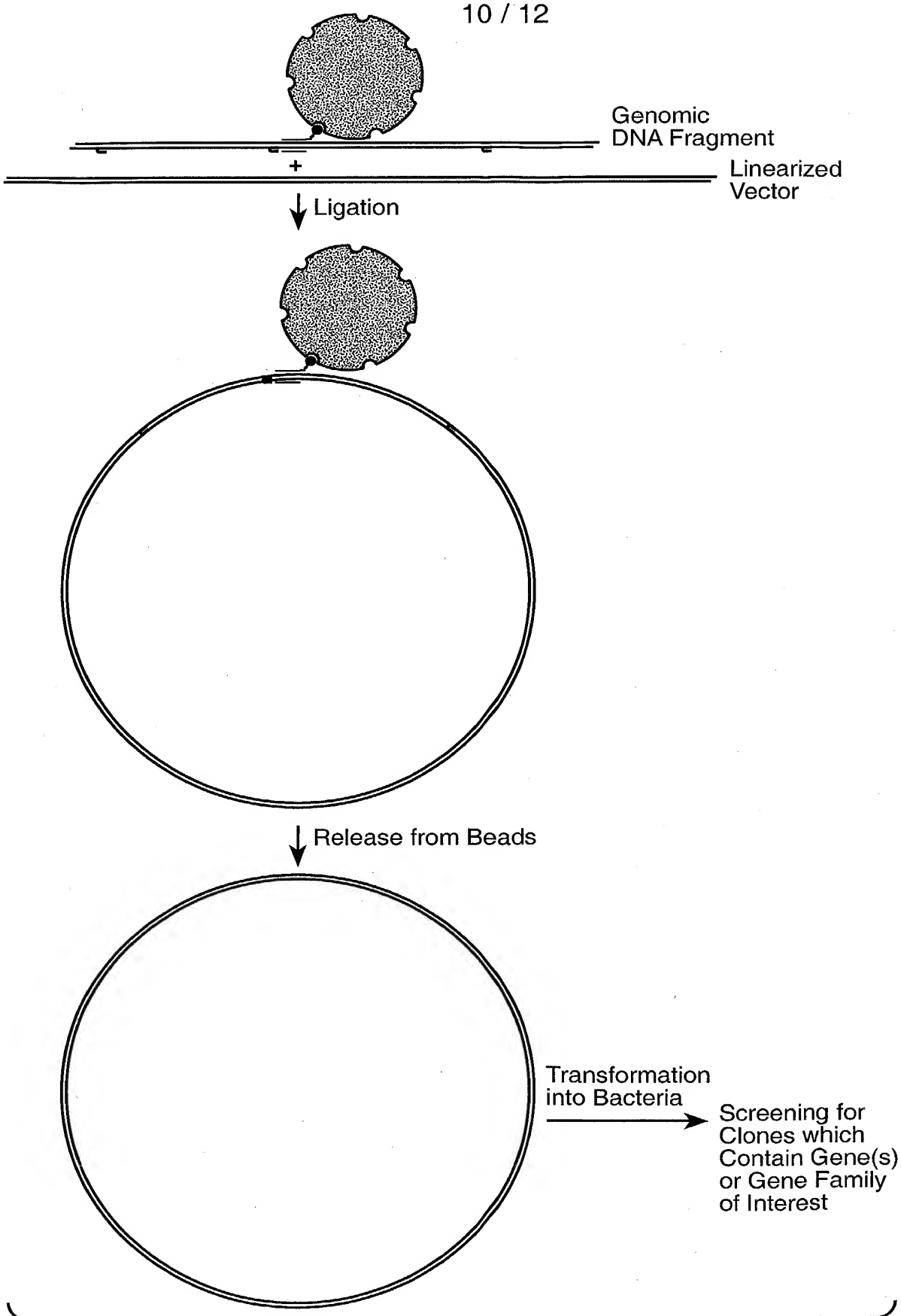
Genomic
DNA Fragment
+
Linearized
Vector
Ligation
Release from Beads
Transformation
into Bacteria
Screening for
Clones which
Contain Gene(s)
or Gene Family
of Interest

FIG._4B

Scheme for Targeting of Human HPRT Gene

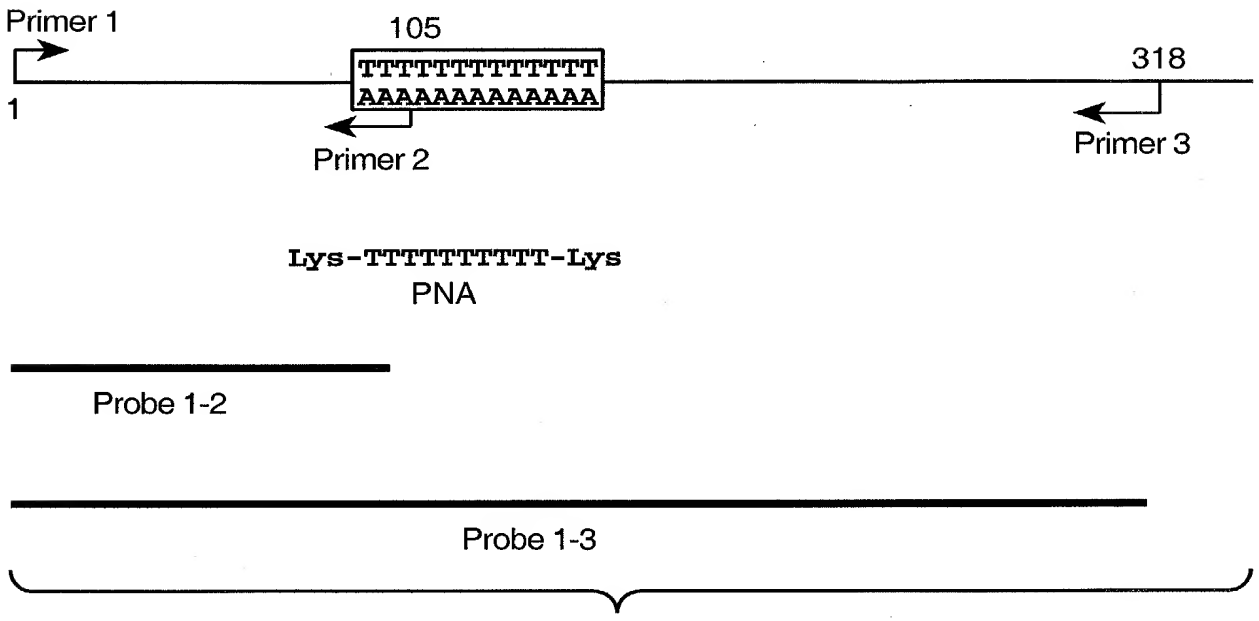

FIG._5A

Targeting of the Human HPRT Gene Fragment with the Probe Adjacent to the PNA Binding Site

Special Target	+				-
PNA	+		-		+
RecA	+	-	+	-	+
Lane	1	2	3	4	5

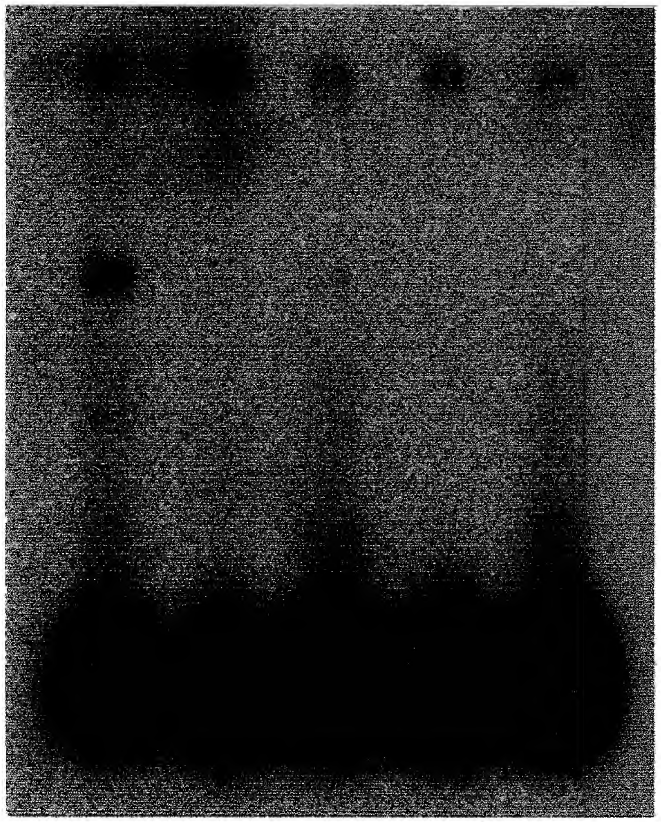

FIG._5B

Targeting of the Human HPRT Gene Fragment with the Probe with the PNA Binding Site Inside It

Special Target	+				-
PNA	+		-		+
RecA	+	-	+	-	+

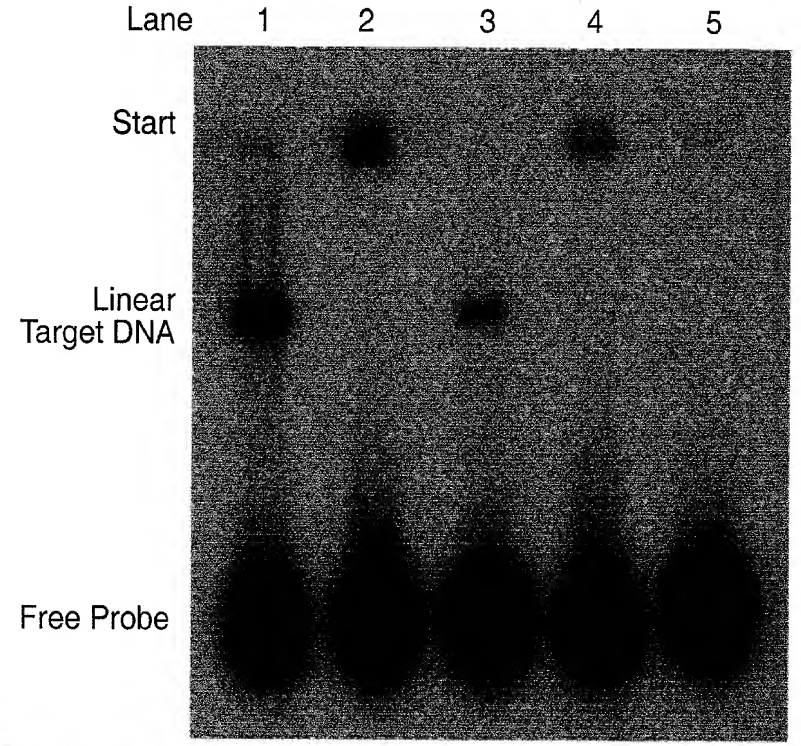

FIG._5C